SHORT COMMUNICATION

A Cautionary Note: Survival of Nymphs of Two Species of Ticks (Acari: Ixodidae) Among Clothes Laundered in an Automatic Washer

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ABSTRACT Host-seeking ticks often remain on clothing of persons returning home from work or recreation in tick habitats, and can pose at least a temporary risk to people and pets in these homes. Laundering clothing has been one of the recommendations to reduce tick exposure. Host-seeking lone star tick, *Amblyomma americanum* (L.), and blacklegged tick, *Ixodes scapularis* Say, nymphs confined in polyester mesh packets, were included with laundry in cold, warm, and hot wash cycles of an automatic clothes washer. Ticks were also placed with washed clothing and subjected to drying in an automatic clothes dryer set on high heat and on air only (unheated). Most nymphs (≥90%) of both species survived the cold and warm washes, and 95% of *A. americanum* nymphs survived the hot wash. At the time of their removal from the washer, *I. scapularis* nymphs were clearly affected by the hot wash, but 65% were considered alive 20–24 h later. Large percentages of nymphs of both species survived hot washes in which two other detergents (a powder containing a nonchlorine bleach and a liquid) were used. All ticks were killed by the 1 h cycle at high heat in the clothes dryer, but with unheated air some nymphs of both species survived the 1 h cycle in the dryer. Given the laundering recommendations of clothing manufacturers and variation in the use automatic clothes washers, laundry washed in automatic washers should not be considered free of living ticks.

KEY WORDS blacklegged tick, *Ixodes scapularis*, lone star tick, *Amblyomma americanum*

BECAUSE OF THEIR ABILITY to transmit disease causing pathogens, ticks are a serious problem to persons working, visiting and living in habitats where ticks occur. Often when a tick is acquired by accident by a person outdoors, it is via contact with the person's clothing, such as shoes, socks and pants (Carroll and Kramer 2001) and, it is not uncommon for persons to return home with ticks on their clothing. If tick infested clothing is removed and left somewhere, such as in a hamper, in the home, the risk of tick bite still exists. More active species can crawl from clothing left in a bedroom to sleeping persons, and other ticks can be contacted by persons handling the used clothing. One way to reduce the risk of being bitten by ticks brought into the home on clothing is to remove clothing worn in the field, and immediately place them in a sealed plastic bag. The bag just confines any ticks until the clothes are washed. The common perception is that once the clothes have been laundered, that is the end of the matter regarding ticks.

Common in many parts of the southern United States, lone star ticks, *Amblyomma americanum* (L.), can transmit the agent causing monocytic ehrlichiosis (Walker and Dumler 1996). The blacklegged tick, *Ixodes scapularis* Say, is becoming increasingly common in parts of the United States, and is the principal vector of the agent causing Lyme disease (Burgdorfer et al. 1982). Larvae, nymphs and adults of both species readily bite humans. The small larvae and nymphs are easily overlooked on a person's skin and clothing. Most cases of human Lyme disease are the result of people being bitten by infected *I. scapularis* nymphs (Lane et al. 1991). The purpose of this study was to ascertain whether lone star ticks, *A. americanum*, and *I. scapularis* nymphs survive typical American laundering practices, involving an automatic washer and a dryer, and thus present a risk of tick bite to persons handling recently washed clothing.

Materials and Methods

Lone star tick nymphs were obtained from the USDA, ARS Knipling-Bushland U.S. Livestock Insects Research Laboratory, Kerrville, TX courtesy of J. M. Pound. Unfed *I. scapularis* larvae were purchased from Oklahoma State University (J. Bowman), Stillwater, OK and allowed to feed to repletion on a rat (in accordance with USDA, ARS Beltsville Area Animal Care and Use Committee, Animal Use Protocol #02-015). Both species of ticks were maintained at ≈22°C, ≈97% RH, and a photoperiod of 16: 8 h (L:D). At the

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time of testing the *A. americanum* nymphs were 4–5 wk in that stage, and *I. scapularis* 2–3 wk as nymphs.

Ten active nymphs (either all *I. scapularis* or all *A. americanum*) were placed in a flat, polyester fabric (25 × 11 mesh/cm) packet (≈2.8 × 6 cm) that had two edges sealed with glue. The opening through which the ticks were inserted was closed with a bulldog clip (Boston Clip, No. 2, Hunt Manufacturing Corporation, Statesville, NC). Two packets each of *A. americanum* and *I. scapularis* nymphs were placed within a larger mesh bag (25 × 12.5 cm). To serve as controls, one packet containing 10 *A. americanum* nymphs and another, containing 10 *I. scapularis* nymphs, were placed in plastic bags each containing a wad of moistened tissue. The control ticks were kept at 20–21°C in the laundry room. Each load of laundry was the same, consisting of three shirts, two pair of jeans, three T-shirts, three pair of socks, two pair of lightweight coveralls, and two pair of heavy coveralls. A Whirlpool (Benton Harbor, MI) super capacity, two speed automatic washer (model LA558T) was used for all the trials. All washes were set for a medium load and a normal cycle (30 min), and one of three temperature settings (cold/ cold, warm/cold, hot/cold). The normal cycle consisted of filling, 4 min wash at high speed, 6 min wash at low speed, draining (no agitation), spinning at high speed, filling for rinse, draining (no agitation), and spinning at high speed with a spray rinse. In a random order, four washes were done for each temperature setting. ≈100 g of powdered detergent (Clout, Costco Wholesale Corporation, Seattle, WA), an amount slightly greater than the manufacturer's recommendation for a moderate size load of wash, was added as water was flowing into the washer. The bag containing the ticks was placed among the clothes in the washer on the side opposite the water inflow, and a little above the vertical midpoint of the pile of clothes in the washer. The temperature of the inflowing water was obtained by placing a thermometer in the stream for ≥90 s. Immediately after completion of the cycle, a thermometer was inserted among the clothing in the washer, the packets of ticks were removed from the washer and the ticks examined for survival. To help determine the condition of the ticks, those that were immobile were removed from packets and exhaled upon or lightly prodded with forceps. Ticks were categorized as alive, dead or moribund (appendages moving, but tick not crawling). These procedures were used to determine the effects of two additional detergents, Tide (a powder with an oxygen bleach, Proctor and Gamble, Cincinnati, OH) and Heavy Duty Ultra (a liquid, Rite Aid Corporation, Harrisburg, PA) on nymphs of both species. Ten nymphs of each species were tested in each of three washes (hot/cold cycle) containing (in accordance with manufacturers' recommendations) ≈100 g of Tide and ≈90 ml of Heavy Duty Ultra. Equal numbers of control ticks were kept and observed as described for the Clout tests.

To obtain some indication of the affect of hot water on nymphs, one packet containing 10 *I. scapularis* nymphs and another with 10 *A. americanum* nymphs were placed in a cup and held in the flow of water entering the washer. Hot water filled and overflowed the cup for 1 min, after which the ticks removed and examined for survivors. An equal number of control ticks kept in plastic bags with wads of moist tissue were similarly examined. This was repeated for three separate wash cycles. Temperature of the water was simultaneously recorded. One packet of nymphs of each species, that had just been removed from the washer and examined, was randomly chosen and placed in the plastic bag with the control ticks. The other packet was placed along with the damp laundered clothes in a gas operated clothes dryer (Whirlpool, model LGQ9558KQ0, Benton Harbor, MI). The bag containing the packets of ticks was placed amid the pile of wet clothes in the dryer, and the dryer operated at high heat for 1 h. The 1 h cycle included a cooling period for the last ≈20 min. Following the same protocol, an additional three groups of nymphs were subjected to cold/cold washes and dried with the clothes at the unheated air setting for 1 h. Immediately after the dryer cycle was completed, the ticks were removed and examined for survivors. The packets were placed in the plastic bags containing the controls and the other nymphs laundered in the same batch. At 20–24 h after the ticks were removed from the washer they were examined and the numbers of living, dead, and moribund nymphs recorded. Only ticks exposed to the Clout detergent were tested in the dryer. To ascertain temperatures inside the dryer, wet loads of the standard wash were dried, the dryer stopped mid cycle and a thermometer placed among the clothes, and repeatedly checked until a maximum temperature was reached. Immediately at the end of the high heat cycle (when the ticks were removed) of the last five dryer treatments, a Thermo Hygro hygrometer/thermometer (Sper Scientific, Scottsdale, AZ) was placed in the dryer on top of the pile of dried clothes, and the dryer door shut. After 2 min the hygrometer/thermometer was retrieved and the RH recorded. Because the distribution of ticks was multinomial (alive, moribund, dead), the data were analyzed using a generalized linear model (Proc Genmod) with a cumulative logit link (SAS Institute 1999).

Results

Immediately after their removal from cold and warm washes, nearly all (≥90%) *A. americanum* and *I. scapularis* nymphs were alive, a survival level no different from untreated control ticks ($P > 0.05$) (Table 1). Survival (95%) of *A. americanum* nymphs immediately after their removal from a hot wash did not differ from control ticks ($P > 0.05$). In contrast, most *I. scapularis* nymphs were clearly affected by the hot wash. The *I. scapularis* were slow to crawl or even move, with many nymphs (31.3%) categorized as moribund (Table 1). Overall 23.8% of the *I. scapularis* nymphs subjected to a hot wash were considered alive upon removal from the washer, but there was considerable variation among the hot washes in numbers of dead (20, 12, 3, 2) and moribund (0, 8, 13, 3) *I. scapu-*

Table 1. Number and percent (in parentheses) of host-seeking *A. americanum* and *I. scapularis* surviving cold, warm and hot washes immediately and 24 h after their removal from the washer

	Number (%) nymphs					
	A. americanum			*I. scapularis*		
	Alive	Moribund	Dead	Alive	Moribund	Dead
Immediately after wash						
Cold	80 (100)	0 (0)	0 (0)	72 (90)	0 (0)	8 (10)
Warm	78 (97.5)	0 (0)	2 (2.5)	80 (100)	0 (0)	0 (0)
Hot	76 (95)	0 (0)	4 (5)	19 (23.8)	25 (31.3)	36 (44.9)
24 h after wash						
Cold	40 (100)[a]	0 (0)	0 (0)	39 (97.5)	0 (0)	1 (2.5)
Warm	38 (95)	0 (0)	2 (5)	38 (95)	(0)	2 (5)
Hot	38 (95)	0 (0)	2 (5)	26 (65)	2 (5)	12 (30)

[a] Half of washed packets used in dryer tests, so n = 40 for 24 h. All control ticks were alive immediately and 24 h after the washes.

laris nymphs. The mean inflowing water temperature was 10.7 ± 0.3°C for cold/cold washes, 29.5 ± 0.3°C for the warm/cold washes, and 50.7 ± 0.3°C for the hot/cold washes. Temperatures measured in the washer among the clothes immediately after the wash cycle ended were 13.6 ± 0.3°C in the cold/cold wash, 15.6 ± 0°C in the warm/cold wash, and 16.3 ± 0.4°C in the hot/cold wash. All *I. scapularis* and *A. americanum* nymphs that were submerged for 1 min in the hot water (50.6°C) that flowed into the washer were alive when removed from the water.

At 20–24 h after the cold and warm washes, survival rates of *I. scapularis* and *A. americanum* nymphs remained high and similar to controls ($P > 0.05$) (Table 1). Similarly *A. americanum* nymphs subjected to a hot wash showed little decline in survival after 20–24 h. However, many *I. scapularis* nymphs that were considered moribund when they were removed from a hot wash were able to walk normally at 20–24 h, and categorized as alive. Nevertheless, significantly fewer ($P < 0.01$) *I. scapularis* nymphs survived than *A. americanum* nymphs. All the ticks (three groups of 10 nymphs of each species) that were submerged in the hot water inflow survived for 20–24 h.

Most nymphs were found alive immediately after the hot/cold washes with Tide (63.3% *I. scapularis*, 96.7% *A. americanum*, $n = 30$ for each species), and with the revival of moribund ticks 90% of the *I. scapularis* nymphs were considered alive 24 h after the wash. Results with the liquid Heavy Duty Ultra were similar with 66.7% of *I. scapularis* and 83.3% of *A. americanum* nymphs alive immediately after the wash, and 80% of *I. scapularis* and 90% of *A. americanum* nymphs alive at 24 h after the wash. Control survival was 96.7–100% for the Tide and Heavy Duty Ultra tests.

All nymphs of both species were dead when removed from the dryer after a 1 h cycle at the high heat setting. In most cases they were severely desiccated, often with appendages missing. Temperatures of 40–42°C were recorded in the dryer midway through the cycle at the high setting, and 19–20°C at the air only setting. The mean RH at end of the high heat cycle was 53.8 ± 7.5%, ranging from 28 to 74%. When unheated air was used in the dryer, 53.3% of the *A. americanum* and 30% of the *I. scapularis* nymphs were alive when removed from the dryer, and 66.7% and 26.7%, respectively, were alive at 20–24 h after being removed from the dryer. Survival of control ticks not placed in the dryer was 100% for *A. americanum* and 99.3% for *I. scapularis* nymphs 24 h after the treatment. Thus, significantly more ticks survived the unheated drying than the heated drying ($P < 0.01$), but significantly ($P < 0.01$) fewer ticks survived the unheated drying than ticks not subjected to drying. Some of the ticks that died in the air only cycle appeared physically damaged, perhaps crushed or pinched because of the weight of the wet clothing that had not dried by the end of the cycle.

Discussion

The high frequency of survival of *A. americanum* and *I. scapularis* nymphs that were subjected to cold and warm washes is cause for wariness in handling laundered clothing that had been worn in tick habitats. Hot washes did not kill *A. americanum* nymphs, and variably harmed *I. scapularis* nymphs, indicating that even more extreme conditions may be needed to rid clothing of host-seeking ticks. The only difference between the two species in their susceptibility to the washes was the greater mortality and morbidity of *I. scapularis* nymphs subjected to the hot washes. The nymphs of *A. americanum* were slightly larger than those of *I. scapularis* and the former are distributed widely in hotter climates (Sonenshine 1993). The variability in survival of *I. scapularis* in hot washes may have been because of where the packets were located at some point the cycle that exposed them to hot water. Short exposures (submersion for 1 min) to the hot water (50°C) did no apparent harm to nymphs of either species. Furthermore, the water temperature in the washer dropped as it contacted the clothing, which was at ambient temperature (20–21°C). However, because water temperature was the only difference among the washes, the mortality of *I. scapularis* in the hot washes was probably caused by exposure to hotter water. Drowning during the relatively short wash cycle is not a likely threat to tick survival. Supporting this assertion is a collection technique used by the author (unpublished data), in which captured *I. scapularis* nymphs were submerged in water in a vial,

and 2–3 h later in the laboratory removed from the water with recovery of all ticks. Although not included in this study, adult American dog ticks, *Dermacentor variabilis* (Say), are the primary vectors of the agent causing Rocky Mountain fever, and another species likely to be brought into homes on clothing (Smith et al. 1946). According to Smith et al. (1946), nymphs of *D. variabilis* survived submersion for a day in fresh and in sea water. In the washer, water did not completely fill the wash chamber, and water was absorbed by the fabrics, so ticks, depending on their location, may not, or only briefly, be submerged in the hottest water. The ticks in this study were confined to recoverable mesh packets, but in actual laundering situations an unknown portion of ticks loose among clothing may be swept down the drain. However, because of their renowned adhamant abilities, ticks, when protected by layers and folds of fabric, may not be readily dislodged by the action of the agitator and water currents. Some washers (the one used in this study) do not agitate the wash during draining. This study was undertaken in part, because the author found a live male *A. americanum* on the agitator of the washer when he was about to remove the wash. Some of the minimal tick mortality observed in this study may have been because of the use of metal bulldog clips to close the packets. Dead ticks were appeared physically damaged or crushed, perhaps by being caught between bulldog clips confined in close proximity in the sock bag.

In the routine laundry practices in the United States, the use of hot water washes is probably limited. Clothing manufacturers recommend cool or cold washes for a wide variety of fabrics and colored garments. Water heater manufacturer recommendations discourage setting water temperatures above 54°C, because of the danger of scalding. To be effective against ticks the water would have to be hotter than that used in this study (≈50°C). There are a few principal manufacturers of clothes washers, who each produce a few models of washer each with options for temperature of water, volume of water, and duration of cycle. Consequently, there are a variety of combinations of conditions from which one can select, but for the purposes of this study a moderate volume of water and "normal" cycle was used. The impact of chlorine bleach on tick survival was not examined in this study, because laundering recommendations for colored clothing generally warn against the use of bleach. The intent of this study was to explore the possibility of tick survival among clothing laundered in an automatic washer. Because ticks were found to survive rather usual laundry conditions, further evaluation of tick survival for the numerous combinations of detergents, bleaches, wash cycles, and washing machines was unnecessary.

The dryer killed all the ticks exposed to a 1 h cycle at high heat, which substantiates the CDC recommendation for washing and drying clothing at a high temperature after being outdoors (CDC 2001). According to the manufacturer, at high heat, the temperature in the dryer reaches ≈68°C, much hotter than the hot wash, and >20°C higher than the temperatures I recorded from near the top of the clothing piled in the dryer. Accompanying the elevated temperatures in the dryer, were the effects of the dryer's exhaust fan that dried the clothes and would tend to lower the relative humidity in the dryer, further compromising the survival of the nymphs. *I. scapularis* is particularly susceptible to desiccation (Curran and Needham 1996). The average RH at end of the high heat cycle was 53.8%, but in one case was as low as 28% RH. Apart from the effects of the heat in the dryer, ticks loose among the clothing, are subjected to prolonged tumbling and if dislodged they could be evacuated by the strong exhaust current. There seems to be risk of physical damage to ticks as well in the dryer, with many of the dead ticks killed during the air only cycle appearing to have been crushed.

Automatic clothes washers and dryers were not designed to kill ticks, and washing tick infested clothing in automatic clothes washers should not be expected to effectively kill ticks. Some ticks may be carried away in the water as it drains from the washer, but risk still remains to a person removing clothes from the washer and handling line or rack dried laundry. Drying clothes in an automatic dryer at high heat for 1 h appears to offer a greater level of certainty of ridding laundry of host-seeking ticks. However, because of the variety and combinations of settings available on washers and dryers, and the state of the maintenance of individual machines, the effectiveness of particular private and commercial machines against ticks is unknown.

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